

**WHAT IS CLAIMED IS:**

1 1. A Lac shuttle vector, comprising:

2 (a) a region which regulates a plasmid copy number,  
3 wherein said region comprises an *E. coli* replication origin  
4 sequence;

5 (b) an eukaryotic gene expression cassette, which  
6 comprises an eukaryotic gene transcriptional promoter  
7 sequence, a multiple cloning site and a transcriptional  
8 terminator sequence, wherein a heterologous gene is inserted  
9 into said multiple cloning site;

10 (c) a lactic acid bacteria plasmid sequence, which  
11 comprises a plus origin of replication, and a nucleic acid  
12 sequence encoding for a protein which relates to the lactic  
13 acid bacteria plasmid replication; and

14 (d) a non-antibiotic resistance selection gene and the  
15 promoter sequence thereof.

1 2. The Lac shuttle vector as claimed in claim 1,  
2 wherein said eukaryotic gene transcriptional promoter is  
3 cytomegalovirus (CMV) promoter.

4 3. The Lac shuttle vector as claimed in claim 1,  
5 wherein said lactic acid bacteria plasmid sequence is the  
6 plasmid of 2.1 kb size isolated from *Lactobacillus plantarum*.

7 4. The Lac shuttle vector as claimed in claim 3,  
8 wherein the protein which relates to the lactic acid  
9 bacteria plasmid replication is Rep A protein containing 317  
10 amino acids.

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1 5. The Lac shuttle vector as claimed in claim 1,  
2 wherein said non-antibiotic resistance selection gene is  $\beta$ -  
3 galactosidase gene.

1 6. The Lac shuttle vector as claimed in claim 5,  
2 wherein the promoter of said  $\beta$ -galactosidase gene is  
3 erythromycin resistance gene promoter.

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1 7. The Lac shuttle vector as claimed in claim 1,  
2 wherein the Lac Shuttle vector comprises the nucleotide  
3 sequence set forth in SEQ ID NO:1 or a complementary  
4 nucleotide sequence thereto, or a degenerate variant thereof.

1 8. The Lac shuttle vector as claimed in claim 1,  
2 wherein the Lac Shuttle vector comprises the nucleotide  
3 sequence set forth in SEQ ID NO:2 or a complementary  
4 nucleotide sequence thereto, or a degenerate variant thereof.

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1 9. The Lac shuttle vector as claimed in claim 1,  
2 wherein the Lac Shuttle vector is selected from the group  
3 consisting of:

4 (a) pCLP7 (having the configuration of restriction  
5 sites in FIG.3, ATCC Accession No. PTA-2661); and

6 (b) pCLP8 (having the configuration of restriction  
7 sites in FIG.3, ATCC Accession No. PTA-2663).

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1 10. The Lac shuttle vector as claimed in claim 1,  
2 wherein the host cell being transformed is a Gram-positive  
3 bacterium, and the endogenous  $\beta$ -galactosidase gene of the

4 host cell is not capable of producing a normal enzymatic  
5 function.

1 11. The Lac shuttle vector as claimed in anyone of  
2 claim 10, wherein the host cell is the mutant of  
3 *Lactobacillus casei* (subsp. *casei*), which is designated Ana-  
4 1 (Lac<sup>-</sup> mutant), (ATCC Accession No. PTA-2662).

1 12. A kit for expression of a heterologous gene,  
2 comprising:

- 3 (a) the Lac shuttle vector as claimed in claim 1;  
4 (b) a host cell which the endogenous  $\beta$ -galactosidase  
5 gene thereof is not capable of producing a normal enzymatic  
6 function; and  
7 (c) an eukaryotic cell.

1 13. A DNA vaccine carrier comprising the Lac shuttle  
2 vector as claimed in claim 1.

1 14. A method for selection of a host cell containing a  
2 vector, comprising:

- 3 (i) introducing into said host cell the Lac shuttle  
4 vector as claimed in claim 1,  
5 wherein the endogenous  $\beta$ -galactosidase gene of said  
6 host cell is not capable of producing a normal enzymatic  
7 function; and  
8 (ii) culturing said host cell transformed in step (i)  
9 under conditions which lactose is the only carbon source.